# 70439 Oxidase Test

Oxidase Test is an important differential procedure which should be performed on all gram-negative bacteria that are to be identified.

**Composition:**
(1 package contains 50 discs)
discs contains N,N-dimethyl-p-phenylenediamine oxalate and α-naphthol

**Directions:**
Take an inoculating loop or a toothpick. Then touch and spread a well isolated colony on an oxidase disc. The reaction is observed within 5 to 10 seconds at 25-30°C. A change later than 10 seconds or no change at all is considered as negative reaction.

**Principle and Interpretation:**
Gordon and McLeod (1) introduced oxidase test for identifying Gonococci based upon the ability of certain bacteria to produce indophenol blue from the oxidation of dimethyl-p-phenylenediamine and α-naphthol. Gaby and Hadley (2) introduced a more sensitive method by using N,N-dimethyl-p-phenylenediamine oxalate where all Staphylococci were oxidase negative.
In presence of the enzyme cytochrome oxidase (gram-negative bacteria) the N,N-dimethyl-p-phenylenediamine oxalate and α-naphthol react to indophenol blue.
Oxidase test is mainly used to differentiate:
1) Oxidase positive *Neisseria* from other gram-negative diplococci.
2) Oxidas positive *Aeromonas hydrophila* from *Escherichia coli* (gram-negative)
3) Oxidase positive *Plesiomonas shigelloids* from *Shigella sonnei* (gram-negative)

*Note:*

1. Do not use stainless steel or nichrome inoculating wires, as false positive reaction may result from surface oxidation products formed during flame sterilization.
2. Growth from media containing dyes is not suitable for testing.
3. Timing is critical (5-10 sec) for interpretation of results.
4. Perform oxidase test on all gram-negative bacilli.
5. Cytochrome oxidase production may be inhibited by acid production and false negative reaction may be given by *Vibrio*, *Aeromonas*, and *Plesimonas* species when grow on a medium containing fermentable carbohydrate e.g. MacConkey Agar (70143). Colonies taken from media containing nitrate may give unreliable results. The loss of activity of the oxidase reagent is caused by auto oxidation which may be avoided by adding 0.1% ascorbic acid (95209).

Reaction within 5 to 10 seconds at 25-35ºC.

| Organisms (ATCC) | Reaction | Color |
|---|---|---|
| *Pseudomonas aeruginosa* (27853) | positive | deep purple blue |
| *Staphylococcus aureus* (25923) | negative | - |
| *Neisseria gonorrhoeae* (19424) | positive | deep purple blue |
| *Escherichia coli* (25922) | negative | - |

The vibrant M, Millipore, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates.
Detailed information on trademarks is available via publicly accessible resources.
© 2018 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.
The life science business of Merck KGaA, Darmstadt, Germany
operates as MilliporeSigma in the US and Canada.

References:
1. J.Gordon, J.W. McLeod, J. Path.Bact., 31, 185 (1928)
2. W.L. Gaby, C. Hadley, J. Bact., 74, 365 (1957)
3. K.J. Steel, J. Appl. Bact., 25, 445 (1962)

**Precautions and Disclaimer**
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

The vibrant M, Millipore, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates.
Detailed information on trademarks is available via publicly accessible resources.
© 2018 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.
The life science business of Merck KGaA, Darmstadt, Germany
operates as MilliporeSigma in the US and Canada.